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For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: METHODS OF PROMOTING THE DIFFERENTIATION OF STEM CELLS

(57) Abstract: A method is described by which stem cells are induced to differentiate into cardiomyocytes; the method comprises exposure for a length of time and at efficacious quantities of a protein of the EGF-CFC family or its derivatives having at least the EGF and CFC domains; or to differentiate into neuronal cells, comprising the exposure to Cripito protein inhibitors. Compositions are described for therapeutic use in treating heart disorders, comprising a therapeutically efficacious quantity of a protein or its derivatives having at least the EGF and CFC domains of a protein of the EGF-CFC family.

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INTERNATIONAL SEARCH REPORT

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A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 C12N5/06 A61K35/34 A61K38/18 A61P9/00 A61K39/395
A61K35/30 A61P25/28

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 C12N A61K C07K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, WPI Data, BIOSIS, EMBASE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	XU CHUNHUI ET AL: "Specific arrest of cardiogenesis in cultured embryonic stem cells lacking Cripto-1" DEVELOPMENTAL BIOLOGY, vol. 196, no. 2, 15 April 1998 (1998-04-15), pages 237-247, XP002294486 ISSN: 0012-1606 cited in the application the whole document	1-12
X	WO 01/68814 A (FIELD LOREN J ; ADVANCED RES & TECH INST (US)) 20 September 2001 (2001-09-20) examples ----- -/--	6-8

☒ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

* Special categories of cited documents:

- *A* document defining the general state of the art which is not considered to be of particular relevance
- *E* earlier document but published on or after the international filing date
- *L* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- *O* document referring to an oral disclosure, use, exhibition or other means
- *P* document published prior to the international filing date but later than the priority date claimed

- *T* later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- *X* document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
- *Y* document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.
- *G* document member of the same patent family

Date of the actual completion of the international search

2 September 2004

Date of mailing of the international search report

17/09/2004

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INTERNATIONAL SEARCH REPORT

International Application No

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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	ADAMSON E D ET AL: "Cripto: A tumor growth factor and more" JOURNAL OF CELLULAR PHYSIOLOGY, vol. 190, no. 3, March 2002 (2002-03), pages 267-278, XP002294487 ISSN: 0021-9541 cited in the application -----	
X	WO 02/26941 A (KOOY DEREK V D ; TROPEPE VINCENT (US)) 4 April 2002 (2002-04-04) page 34, line 4 - line 31 -----	13-22
E	WO 2004/053084 A (MCLEAN HOSPITAL CORP) 24 June 2004 (2004-06-24) page 5, line 5 - line 8 page 43 - page 48 -----	13-22
A	PICCOLO S ET AL: "The head inducer Cerberus is a multifunctional antagonist of Nodal, BMP and Wnt signals" NATURE, vol. 397, no. 6721, 25 February 1999 (1999-02-25), pages 707-710, XP002294488 ISSN: 0028-0836 cited in the application -----	
A	BALDASSARRE G ET AL: "Transfection with a cripto anti-sense plasmid suppresses endogenous cripto expression and inhibits transformation in a human embryonal carcinoma cell line" INTERNATIONAL JOURNAL OF CANCER, vol. 66, no. 4, 16 May 1996 (1996-05-16), pages 538-543, XP008006513 ISSN: 0020-7136 -----	
X,P	PARISI S ET AL: "Nodal-dependent Cripto signaling promotes cardiomyogenesis and redirects the neural fate of embryonic stem cells." JOURNAL OF CELL BIOLOGY, vol. 163, no. 2, 27 October 2003 (2003-10-27), pages 303-314, XP002294489 ISSN: 0021-9525 the whole document -----	1-22

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Box No. I Nucleotide and/or amino acid sequence(s) (Continuation of item 1.b of the first sheet)

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application and necessary to the claimed invention, the international search was carried out on the basis of:
 - a. type of material
 - ☒ a sequence listing
 - ☐ table(s) related to the sequence listing
 - b. format of material
 - ☒ in written format
 - ☒ in computer readable form
 - c. time of filing/furnishing
 - ☐ contained in the international application as filed
 - ☐ filed together with the international application in computer readable form
 - ☒ furnished subsequently to this Authority for the purpose of search
2. ☒ In addition, in the case that more than one version or copy of a sequence listing and/or table relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.
3. Additional comments:

INTERNATIONAL SEARCH REPORT

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Box II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
2. ☐ Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☒ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. claims: 1-12

Method to induce stem cell differentiation into cardiomyocytes using a protein of the EGF-CF family; differentiated stem cells, composition and use thereof. A composition for treating heart disorders comprising a protein of the EGF-CF family.

2. claims: 13-22

Method to induce stem cell differentiation into neuronal cells using a Cripto inhibitor; differentiated stem cells, composition and use thereof.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/IT2004/000133

Patent document cited in search report		Publication date	Patent family member(s)	Publication date
WO 0168814	A	20-09-2001	AU 4355101 A CA 2402245 A1 EP 1265987 A2 JP 2004504807 T WO 0168814 A2	24-09-2001 20-09-2001 18-12-2002 19-02-2004 20-09-2001
WO 0226941	A	04-04-2002	AU 9358601 A WO 0226941 A2 CA 2424062 A1 US 2002164791 A1	08-04-2002 04-04-2002 04-04-2002 07-11-2002
WO 2004053084	A	24-06-2004	WO 2004053084 A2	24-06-2004